PROGRESS OF JAPANESE BEETLE INVESTIGATIONS

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Investigations of the Japanese beetle (*Popillia japonica* Newman) were begun by the Bureau of Entomology in 1917, following the discovery of the insect near Riverton, N. J., in August 1916. At first, efforts were centered chiefly upon an attempt to exterminate the infestation and to acquire a general knowledge of the life history and habits of the insect, but as soon as it became evident that the beetle could not be eradicated, the investigation was directed toward control and reduction of damage. With this objective the following lines of research have been carried on: (1) Obtaining a full and intimate knowledge of the insect's habits and reactions to its environment; (2) development and perfection of measures to prevent material damage by the insect in any of its stages to economic plants and crops; (3) development of practical and economical methods for insuring freedom from infestation of commercially grown nursery stock and agricultural products, to prevent widespread distribution of the insect throughout the United States; and (4) introduction of predacious and parasitic enemies of the beetle from the Orient and their dissemination throughout the infested areas.

It is the purpose of this paper to review briefly some of the results accomplished by the investigation and to refer to the major lines of study under way at the present time.

A full account of the life history and habits of the beetle under conditions obtaining in the older infested area has been published (*17*)[1] The development of the beetle in the more recently invaded areas is being studied as opportunity is offered, to observe its reaction to the different environment conditions.

The probable ultimate distribution of the beetle in the North American Continent is a matter of much interest. While as yet

[1] Italicized numbers in parentheses refer to Literature Cited.

no certain prediction can be made of its ultimate range, studies carried over a period of years of the climatic adaptability of the insect, supplemented by a critical comparison of the outstanding climatic features of the United States with those of Japan, suggest the probability that the Japanese beetle will find climatic conditions in general adapted to its permanent establishment in those sections of the eastern half of the United States where the normal temperature and precipitation most closely approach those of Japan. Generally speaking, this region extends in the Eastern States from the Canadian border south to central South Carolina and northern Georgia, and in the Central States from the southern peninsula of Michigan, southern Wisconsin, and central Iowa to northern Alabama, Louisiana, and northeastern Texas. A combination of the low winter temperature, normal to the northern interior, with an absence of snow, would possibly preclude permanent colonization by the beetle in the region west of the Great Lakes. On the other hand, no obvious barrier exists to the ultimate southward extension of its range to Florida and the entire Gulf coast, although in the light of certain facts in the life cycle of the insect as influenced by temperature and summer rainfall, there exists the bare possibility that its spread in the extreme southern sections of this country may prove difficult than would be antecedently expected.

The investigations relating to the development of control measures may be divided conveniently for the purpose of this discussion into three phases, as follows: Control of the adult beetle; control of the immature stages; and methods for growing, handling, or treated nursery stock and agricultural products to insure their freedom from infestation prior to the shipment of these commodities in ordinary commerce.

Control of the adult beetle. During the early years of the investigation it was noted that certain plants were especially attractive to the beetle, and the outstanding attractiveness of geraniol was discovered. This positive attraction of geraniol was utilized in the development of the first beetle trap in 1924, which used a combination of geraniol and eugenol as the attractant. Studies have been continued since that time to improve the effectiveness of the trap. Particular attention has been directed toward the structural features of the trap in order to increase its efficiency and reduce the cost of manufacture. Studies have also been made to determine to what extent the color of the trap influences its efficiency and to determine the best types or combinations of bait. Of the large number of colors and color combinations tested, it has been definitely established that traps painted green and white are superior to those of any other color. Extensive studies of geraniol have been made (*25*), as a result of which standard specifications have now been drawn up for a much cheaper grade of geraniol than heretofore recommended, but having equal attractiveness. It has recently been found that the addition of phenylethyl alcohol to the geraniol-eugenol combination still further increases the attractiveness of this bait without materially increasing the cost. The solid bait previously recommended, in which the attractants were mixed with bran or other similar materials, has been replaced by the geraniol-eugenol combination in liquid form, dispensed by means of a wick, or vaporized from cakes of pumice or clay which have been impregnated with the attractants. The type of trap now recommended, with the improved bait mixture (*23*), will catch approximately 40 times as many beetles under the same operating conditions as the original trap. Public service patents (Nos. 1,968,953 and 1,968,954) covering two of the latest types of beetle traps have been granted to F. W. Metzger, of the laboratory staff.

It was early recognized that the Japanese beetle is repelled to a large extent by the presence of many toxic and nontoxic white materials on fruit and foliage. The use of lead arsenate with a suitable sticker, such as fish oil or ordinary wheat flour, has been recommended for a number of years for the protection of late ripening apples, peaches, and other tree fruits and the foliage of ornamental trees and shrubs, but because of the residue remaining at the time of harvest arsenicals should not be used on early ripening fruits or under other conditions where poisonous residues would be objectionable.

Many materials have been tried, either alone or in combination, as substitutes for lead arsenate for fruit and foliage protection (*4, 7*). It has been found that derris containing 4 to 5 per cent of rotenone is a definite repellent to the Japanese beetle (*12*), the repellent action appearing to be due primarily to the rotenone and deguelin content of the material, although neither of these constituents is any more repellent than derris when used alone. However, derris decreases rapidly in effectiveness upon exposure to sunlight and is readily washed from the fruit and foliage by rain, and even by heavy dews. The emulsified residue from rosin stills has been found to be a cheap and effective sticker for use with derris, and it does not accelerate the decomposition of derris in sunlight (*16*). The use of the derris and rosin residue is now being recommended as a repellent spray for early ripening peaches (*14*), although the application must be repeated at weekly intervals during the height of the beetle season to obtain satisfactory protection. This development is of particular importance, as it is the first time that it has been possible to recommend a material for the protection of early ripening peaches which does not leave any objectionable residue on the fruit at the time of harvest. The results of studies now in progress are such as to warrant the belief that the decomposition of the derris can be materially retarded so that the number of sprays required to give adequate protection can be decreased.

It has also been known for several years that applications of hydrated lime afford a considerable degree of protection to fruit and foliage, but the poor adhesive quality of this material makes its extensive use impractical. To overcome this difficulty, studies have been made of different oils, gums, and other materials as stickers for lime, and it has been found (*24*) that the addition of aluminum sulfate to hydrated lime forms a spray solution which leaves a residue on the foliage that is very repellent to the beetles throughout the entire season. This cheap, nontoxic repellent spray is now being recommended (*14*) for use in commercial apple orchards and on ornamental trees and shrubs growing under conditions where the use of arsenical sprays is not desirable.

In commercial greenhouses in the generally infested areas, the adult beetle does considerable damage to roses by emerging during the winter months and feeding on the buds and blooms. A method has been developed for applying lead arsenate to the soil of the beds in the greenhouses, which destroys the larvæ without impairing the quality or quantity of the plants and blooms (*22*). Further experience with this method shows that it is possible to maintain a practically complete grubproof condition in the treated houses for at least two years, and possibly longer.

Japanese beetles, when present in large numbers, have caused considerable injury to certain crops, such as sweet corn, asparagus, and rhubarb. It has been found that injury to sweet corn can be reduced by dusting with 300-mesh hydrated lime at the rate of 100 pounds to the acre, applying the dust directly upon the developing silk. Preliminary studies have shown that asparagus brush and rhubarb can be protected to some degree by the application of lime and aluminum sulfate. Additional data are necessary, however, before unqualified recommendations of this method can be made.

Control of immature stages. In areas where the beetle population is very dense, larval populations as high as 40 to 50 per square foot are not infrequent and often cause extensive damage to turf as well as to various crops.

In connection with the investigations to find means of destroying larvæ in the soil, it is necessary to determine rather accurately the average concentration of larvæ in a given field. It has been found that the most accurate estimate can be made with the least labor by examining 1 percent of a given area, using 1 square foot as the unit for examination (*11*). The error of the estimate is influenced by the density of the population and the proportion of the field examined.

Larvæ have proved to be serious pests in cultivated turf of lawns, cemeteries, parks, and golf courses. They feed on the roots of the grass immediately below the surface and when in large numbers will cause injury ranging from 50 percent to total destruction. It has been found that the density of a larval population sufficient to cause damage to turf is not always the same, but is dependent on the type of grass, the condition of the soil, the amount of moisture, the availability of plant food, and other factors going to make up a favorable environment. The most satisfactory treatment for the protection of turf is the application of lead arsenate at the rate of 10 pounds to 1,000 square feet of turf area (*5, 15*). This treatment is now common practice throughout the generally infested area. The permanence of the treatment will depend upon many factors, but in general the turf can be kept immune from injury for at least five years by one application of the lead arsenate at the recommended rate.

In connection with the studies on the application of lead arsenate to soil in nurseries and to turf, an extended study has likewise been made of the rate of penetration into and movement of the arsenate through the soil and the effect of various soil types and conditions upon the poison (*10, 13*). In general, it has been found that, under conditions where leaching is a negligible factor, the various arsenates gradually lose their effectiveness in killing the larvæ in the soil. This decrease in effectiveness can probably be attributed to the slow conversion of the arsenic into a form that is not toxic to the larvæ. The effectiveness of lead arsenate as an insecticide varies in different types of soil, the variation being correlated principally with the amount of water-soluble phosphates, ammonia, and magnesium present in the soils. The pH of the soil and the water-soluble manganese, calcium, potash, chlorides, and nitrates appear to have little influence on the insecticidal action. Further studies have shown that the light sandy soils have practically no power to fix arsenates and that the arsenic is gradually lost from the surface layers. In the heavier clay loams and silt loams there appears to be a definite tendency for arsenic to accumulate in the surface layers and to become fixed. There is a wide variation in the susceptibility of different plants to arsenic in the soil. Some plants are readily injured, whereas others appear to be quite tolerant. The age of the plant also seems to be a factor. The effect of arsenic on a plant is governed by the concentration of soluble arsenic in the soil rather than by the total amount of arsenic present. The use of such stomach poisons as lead arsenate cannot be recommended for destruction of the larvæ in the soil in which truck or vegetable crops are being grown because of the absorption of the arsenic by the plants.

Studies on the effectiveness of cultural practices for control of the larvæ have shown that an average reduction of 28 percent can be obtained by the usual plowing and disking with ordinary cultural equipment. This reduction, however, is generally not sufficient to prevent extensive damage to crops in heavily infested areas. It has been found, in cooperation with the Bureau of Agricultural Engineering of the United States Department of Agriculture, that implements of the roto-cultivator type will effect a 70 to 90 percent reduction in the larval population. This type of equipment offers promise for control, and further studies are being continued along this line.

Methods for growing, handling, or treating nursery stock and agricultural products to insure their freedom from infestation prior to shipment. Nursery stock and many agricultural products are commonly grown under conditions in which it is impractical to prevent them from becoming infested. In order that these commodities may be shipped to points outside the regulated areas in compliance with the requirements of the Japanese beetle quarantine, various methods of destroying infestation have been developed. The carrying out of these methods has been accepted as a basis for certification for such movement (*27*).

It has been found that submersion in hot water at a temperature of 112° F. will destroy the infestation in certain perennial plants (*6*). Dips of carbon disulfide emulsion have also been found effective in this connection and are useful for the treatment of individual trees and other nursery stock in small quantities in the nursery rows. Carbon disulfide (*9*) and naphthalene (*8*) have been found effective for the fumigation of potting soil, compost, manure, and other similar materials which are used for growing plants under conditions where the soil is protected from reinfestation. Paradichlorobenzene has recently been found to be effective as a fumigant for destroying the larvæ in soil about the roots of azaleas, and experiments are being continued to determine whether the treatment can be applied safely to other plants.

The carbon disulfide field treatment was found to be inadequate for treating the large blocks of evergreen stock commonly grown in commercial nurseries within the infested area. A practical procedure for destroying infestation of larvæ in the field under these conditions consists in applying lead arsenate prior to July 1 at the rate of 1,500 pounds per acre, and working it uniformly into the soil to a depth of 3 inches. Plots treated in this manner are free from infestation from October 1 until June

15. Treated plots can be maintained indefinitely free of infestation by analyzing the soil for arsenic each spring and adding prior to July 1 the quantity of lead arsenate necessary to restore the arsenical content of the soil to the required concentration. This procedure has been accepted as an approved method for certification (*27*), and is extensively used by commercial nurseries producing large quantities of field-grown stock.

Blueberries, blackberries, raspberries, and other fruits become infested with adult beetles during the process of harvesting and packing for shipment; bananas are infested during their transfer from boats to refrigerator cars. Methods have been developed for fumigating these fruits with calcium cyanide, liquid hydrocyanic acid, carbon disulfide, or ethylene oxide (*26*). Studies are being continued to determine the possibility of applying these treatments to other agricultural commodities.

The possibility of biological control of the Japanese beetle through the agency of predacious and parasitic enemies has been given a great deal of attention, with respect both to native species of parasites or predators normally attacking white grubs and to those species known to attack the beetle in its native habitat, the Orient.

Native insect parasites and predators of white grubs appear to play only a minor rôle in the control of the Japanese beetle in the general Philadelphia area, in spite of the fact that the beetle has been abundant in this area for many years. With the exception of *Tiphia intermedia* Mall., which parasitizes sporadically only a small fraction of 1 percent of Japanese beetle larvæ, no native tiphiids or scoliids have been observed attacking *Popillia*. The predatory groups, such as the carabids, therevids, tabanids, asilids (*18*), and the formicids, prey upon *Popillia* larvæ when contact is made. However, the normal population of these predators in the present areas of infestation is not sufficient to cause any marked decrease in the *Popillia* population, nor has there been observed any marked increase in the population of these predators due to the increased food supply, as represented by the presence of *Popillia* larvæ in great abundance.

On the other hand, the possibility of a reasonable degree of biological control of the beetle in the future through the agency of its introduced parasites presents a much more helpful picture. The status of parasitic control of *Popillia* in the Orient has been intensively studied (*2, 3*), and to date some 17 species have been imported and liberated in the generally infested area. Of these, however, only five species and one racial form, representing two orders, the Diptera and the Hymenoptera, are known to be definitely established.

The dipterous group, which includes *Centeter cinerea* Ald., *Dexia ventralis* Ald., and *Prosena siberita* Fab., as a whole has not proved to be so promising as had been anticipated. These species are but feebly established, owing in part at least, to climatic differences between their old and new environments, changes in the life cycle of the host, and lack of necessary alternate hosts (*20*). *Centeter* is at present distributed over an area of about 252 square miles in the center of the beetle-infested territory. However, it is not entirely synchronized with its host within this area, and the percentage of parasitization, while high at some points at the beginning of the season, drops to a negligible point when the host appears in the field in abundance. Experiments with southern Japanese strains of *Centeter* to improve synchronization with its host have given negligible results in the latitude of heavy beetle infestation. *Dexia* is still represented by only one feebly established colony near Haddonfield, N. J., where a low, moist soil holds a small fraction of the host larvæ through the critical summer season, thus making them available for the second, or summer, generation of the parasite. Laboratory experiments, however, have shown that *Dexia ventralis* will attack native *Phyllophaga* larvæ, and attempts have been made to establish it in an area in Illinois heavily infested with *Phyllophaga* species. Colonies of *Dexia* have been released near Elkton, Md., in an area inhabited by both *Popillia* and native *Phyllophaga.* While as yet no recoveries of *Dexia* have been made at either of these points, the failure to make recoveries does not necessarily indicate that the species has not survived, as sufficient time may not have elapsed for the survivors to have increased to a point where they can be detected. *Prosena siberita* was originally recovered in 1926, but has since remained in a feeble state of establishment in the Moorestown, N. J., area. This species is handicapped by having only a single generation per year, which is not entirely synchronized with the cycle of *Popillia* in the present area of heavy infestation. It may be more useful in northern areas where *Popillia* will have a two-year cycle, and releases will be made in such areas when the host population becomes sufficiently extensive to warrant liberation.

The hymenopterous parasites are represented by the genus *Tiphia,* of which two species and one racial form are now well established, all larval parasites of *Popillia japonica. Tiphia popilliavora* Roh. has been colonized with locally collected material since 1927 (*21*), and to date colonies have been liberated at 513 points in the generally infested area. Of these, 134 liberations were made during the 1935 season. During 1932-33 a survey of 194 of the points of liberation showed that colonies were established at 114 points, representing a 59 percent establishment. Many of the recovered colonies have built up extensively and spread over considerable areas. For several seasons collections have been made from some of the stronger colonies for field liberation, 13,400 females having been collected from two colonies during the summer of 1935.

It has been observed that the *Tiphia popilliavora* population fluctuates considerably at irregular periods, owing to the seasonal fluctuations of its host. A late emergence of *Popillia* throws the predominance of early second-instar host larvæ into the active oviposition period of the parasite, thus creating a less favorable condition for parasite development, as early second-instar host larvæ are less favorable for the parasite than more mature larvæ. To overcome this difficulty, racial strains of *Tiphia popilliavora* having a marked later seasonal period of activity have been brought in from the Orient. A strain from Chosen (Korea), which normally appears a month or more later than the present established Japanese strains, was liberated at two points in 1934 and recoveries were made from both in 1935; additional releases were made during 1935 at nine points. Five colonies of a second group of late *Tiphia popilliavora* strains from the general vicinity of Yokohama, Japan, were also released during 1935, but it is too soon to determine the results.

The other *Tiphia* species, *T. vernalis* Roh., which was first established in 1926 from material imported from Chosen, has also been successfully colonized (*1*). To date (January 1936) liberations have been made at 493 colony centers, for the most part within the area of heavy beetle infestation, and all but 108 of these were from material collected from the earliest established colonies. The extent of establishment is indicated by the fact that 154 colonies of 100 females each were placed in the field during 1934 and 141 colonies in 1935, all field collected from established local colonies. Scouting has shown that about 53 percent of the colonies released are established. A survey made during the latter part of June 1935, at a colony liberated in 1931, showed that, with a host population of 5.2 larvæ per square foot, the parasitization was 67 percent[2]. While this is an unusually high degree of parasitization and is probably far above the normal, it is indicative of the fact that *Tiphia vernalis,* when properly synchronized with its host, should be an effective parasite of *Popillia.*

One of the more important phases of the biological control investigations now in progress is the study of the relationship of soil micro-organisms to the larval population of the Japanese beetle, undertaken in cooperation with the New Jersey Agricultural Experiment Station. The purpose of the study is to determine what organisms in the soil have a definite relationship to the immature stages of the beetle, the extent to which this relationship influences the seasonal fluctuation of the larval population, and the practical utilization of any of these organisms in the reduction of infestations on a large scale.

Four groups of diseases causing mortality of Japanese beetle larvæ have thus far been encountered: (1) The "white" group, probably of bacterial origin, characterized by the whiteness of infected larvæ and the milky appearance of their blood, with the bodies becoming brown after death; (2) the "black" group, in which the bodies of diseased larvæ become brown or black; (3) the fungus group; and (4) the nematode group, of which several apparently distinct species have been found attacking larvæ (*19*). The white group now appears to be the most important, the mor-

[2] Average of 200 square-foot diggings in a plot of 10,000 square feet, representing an examination of 2 per cent of the total area of the plot.

tality of larvæ from this cause being higher than with any of the other types of disease. There are two, and possibly three, similar yet distinct organisms involved in diseases of the white group, and because of the milky consistancy of the blood of affected larvæ these diseases are spoken of as ''milky'' diseases. They are present in the larval population throughout the year, but reach their peak in June in mature larvæ just prior to pupation. At this time approximately 25 percent of the larvæ at certain stations under observation in 1935 were diseased. Milky diseases are infectious and in the field are transmitted by organisms left in the soil by larvæ killed by disease. These diseases are present at most of the places longest infested by the Japanese beetle, but they have not been found at several places more recently infested. Studies are now under way to determine the feasibility of introducing these diseases at points where they do not now occur, and of their utilization in large-scale reduction of larval populations.

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